

Serial No. 10/612,298

Attorney Docket No. 45240-105719

Exhibit B

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

<i>Group.</i>	1639	}
<i>Confirmation No.:</i>	5133	}
<i>Application No.:</i>	10/612,298	}
<i>Invention:</i>	PEPTIDES COMPRISING AROMATIC D-AMINO ACIDS AND METHODS OF USE	}
<i>Applicant:</i>	Byron Anderson	}
<i>Filed:</i>	July 2, 2003	}
<i>Attorney</i>		}
<i>Docket.</i>	45240-105719	}
<i>Examiner:</i>	Christopher M. Gross	}

DECLARATION UNDER 37 C.F.R. § 1.132

Commissioner for Patents
P O. Box 1450
Alexandria, VA 22313-1450

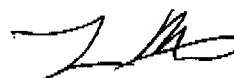
Sir:

1. I, Lenore M. Martin, am not a co-inventor of the patent application captioned-above, but I have made many combinatorial libraries by SPPS in my own laboratory. I am currently an Associate Professor of Cell and Molecular Biology at the University of Rhode Island. My Curriculum Vitae is attached.
2. I have read the patent application captioned above.
3. I understand that the examiner does not believe that Dr. Anderson describes combinatorial libraries of D-peptides in his patent application, where the length of each D-peptide is from three to seven D-amino acid residues, and where at least 68% of the D-peptides have at least 3 aromatic D-amino acid residues – the rest of the positions being either D-alanine or glycine or – if there is room, combinations of both.

4. I understand that the examiner suggests that a person familiar with combinatorial libraries, such as me, could only make a pentapeptide library that satisfies the requirements in paragraph 3 in my Declaration, and not a library of different lengths of D-peptides, e.g., hexapeptides.
5. With the information in Dr. Anderson's patent application, my knowledge, and in accordance with the information in Lebl et al., "One-Bead-One-Structure Combinatorial Libraries," cited in the patent application, I could make a library of 3-7 amino acids where at least 68% of the D-peptides have at least 3 aromatic D-amino acid residues.
6. In split and mix synthetic strategies for combinatorial peptide libraries, the goal is to ensure that all building blocks used to construct the library (in this case D-amino acids such as Phe, Tyr, Trp, Ala, and Gly) are randomly distributed throughout the resulting sequences without bias. This result is ensured by the technique of splitting the mixtures prior to each amino acid coupling step, which avoids the possibility that variations in amino acid coupling reaction kinetics will skew the % incorporation toward any individual amino acid used to construct the library. The result here is that the overall % incorporation of any group of amino acids (such as aromatics) in the resulting peptides can readily be predicted (or controlled) by ascertaining the exact number of coupling reactions in the synthetic sequence that are performed with one of the three aromatic building blocks.
7. For instance, if there are five different building blocks to be used in a random combinatorial synthesis of a peptide library and three of these building blocks are aromatic, then three of nascent peptide mixtures will be coupled to one of the aromatic building blocks during each of the peptide chain elongation steps, with the result that 3/5 of the amino acids incorporated into the library at any chain elongation step will be aromatic. So, after one amino acid is added, 60% of the peptides will contain 1 aromatic amino acid, after the second coupling, 36% will contain 2 aromatics and 60% of the total will contain at least 1 aromatic. After only 3 peptide chain extensions, 21.6% all peptides will already contain the 3 aromatics needed for biological activity, and the percentages of potentially active sequences will continue to grow at each chain elongation step. Once the threshold of 68% aromatic amino acids is crossed, it does not really matter how many additional rounds of coupling are employed.

8. I also believe that Dr. Anderson's combinatorial library fills a need in the field of study of biologically active peptides, because targeted combinatorial libraries, such as those described by Dr. Anderson, derived from a limited subset of at least 3 D-aromatic amino acids (plus D-Alanine and Glycine if there is room), have been shown to yield more productive hits than do combinatorial libraries of random composition.
9. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully submitted,



Name: Lenore Martin, PhD

Title: Associate Professor of Cell &
Molecular Biology, URI

Date: August 28, 2009

Provide the following information for the key personnel and other significant contributors. Follow this format for each person. **DO NOT EXCEED FOUR PAGES.**

Provide the following information for the key personnel and other significant contributors. Follow this format for each person. **DO NOT EXCEED FOUR PAGES.**

A. Positions and Honors.

Other Experience

2005-2007 EPSCOR Louisiana, In-Depth Mail reviewer, Research Competitiveness Subprogram (RCS).

Honors

B. Selected peer-reviewed publications (in chronological order).

Ph.D. dissertation, Orville L. Chapman & Juli Feigon Advisors, David S. Sigman & Frank Anet signatories.

- Martin, Lenore M.; Merrifield, R.B. (1992) "Synthesis and Characterization of Immunoglobulin Variable Region Heavy and Light Chain Fragments" In *Peptides: Chemistry and Biology*; Smith, John A.; Rivier, Jean E. Eds.; ESCOM: Leiden; pp. 849-50.
- Martin, Lenore M.; Rotondi, Kenneth S; Merrifield, R.B. (1994) "McPC603 VH(1-115): Synthesis and Folding of Immunoglobulin Variable Regions", In *Peptides: Chemistry and Biology*; Hodges, R.; Smith, John A.; Eds.; ESCOM: Leiden; pp. 745-47.
- Martin, Lenore M.; Rotondi, Kenneth S; Merrifield, R.B. (1994) "Antibody Binding Constants by Capillary Electrophoresis", In *Peptides: Chemistry and Biology* Hodges, R.; Smith, John A.; Eds.; ESCOM: Leiden; pp. 249-51.
- Martin, Lenore M. (1996) "Facile Reduction in the Synthesis of Phosphorylcholine Affinity Columns" *Tetrahedron Letters*, **37**(44); pp. 7921-24.
- Martin, Lenore M. (1996) "Antibody Fragment Separations by Capillary Zone Electrophoresis" *Journal of Chromatography B*, **675**; pp. 17-25.
- Martin, Lenore M. (1996) "The Use of Ion-Pairing Reagents Improves the Separation of Hydrophobic Peptides by Capillary Electrophoresis" In *Peptides: Chemistry, Structure and Biology*; Kaumaya, P.T.P.; Hodges, R.; Eds.; Mayflower: England; pp. 144-45.
- Li, Chunze; Martin, Lenore M. (1998) "A Robust Method for Determining DNA Binding Constants Using Capillary Zone Electrophoresis" *Analytical Biochemistry*, **263**; pp. 72-78.
- Martin, Lenore M., Hu, Bi-Huang, and Li, Chunze (1998) "Enantiomeric Structural Libraries of Unnatural Peptides which Bind DNA and Alter Enzymatic Activity" *Peptides: Frontiers of Peptide Science*, Tam, J.P. and Kaumaya, P.T.P. eds.), Kluwer Academic Publishing: Dordrecht, Netherlands; pp. 122-23.
- 1999 Hu, Bi-Huang; Martin Lenore M. "Capillary Electrophoresis of Peptides and Proteins", in *High-Resolution Chromatography: A Practical Approach*, Millner, PA ed. IRL/Oxford University Press pp. 77-116.
- Martin, Lenore M. and Hu, Bi-Huang. (1999) "Thiazole and Oxazole Building Blocks for Combinatorial Synthesis" *Tetrahedron Letters*, **40**; pp. 7951-53.
- Martin, Lenore M. "Design of Biomimetic Peptides for Antibiotic Use" (1999) In *Maritimes: Environmental Biotechnology*, **41**(2) Bradley, T. Ed. URI Office of Marine Programs, Summer; pp. 19-21.
- Martin, Lenore M.; Messmer, Bradley T; and Thaler, David S. (2000) "Synthetic Peptide Analogs Compared with Phage Display" in *Peptides for the New Millennium* Fields, Gregg B., Tam, James P., and Barany, George eds., Kluwer Academic Publishers, Dordrecht, The Netherlands, pp. 172-73.
- Hu, Bi-Huang and Martin, Lenore M. (2000) "Design, Synthesis and Antibacterial Activity of a Peptidomimetic Library" in *Peptides for the New Millennium* Fields, Gregg B., Tam, James P., and Barany, George eds., Kluwer Academic Publishers, Dordrecht, The Netherlands, pp. 746-47.
- Martin, Lenore M. Elsaid, Khaled A.; Dorrington, Tarquin; and Gómez-Chiarri, Marta. (2002) "Synthetic Studies on the Antimicrobial Activity of Pleurocidin" In *Peptides: The Wave of the Future* Houghten, Richard A.; Lebl, Michal; Fields, Gregg B.; & Barany, George eds., Kluwer Academic Publishers, Dordrecht, The Netherlands, pp. 475-476.
- Weng, Xiochun, Lutz Hamel, Martin, Lenore M., Peckham, Joan (2005) "A Genetic Algorithm for Energy Minimization in Bio-Molecular Systems" *IEEE Proceedings of the Congress on Evolutionary Computation*, ISBN 0-7803-9363-5, 8 pages.
- W. Lewis Collier, Lenore M. Martin, and Rik van Antwerpen (2007) "Computer-vision determination of 3-D geometric parameters of LDL particles via cryogenic transmission electron microscopy" *Journal of the Society for Information Display* **15**/6: pp. 391-398.
- W. Lewis Collier, Lenore M. Martin, and Rik van Antwerpen (2007) "Accuracy Limits for Efficiently Determining Shape and Size of Low-Density Lipoprotein Macromolecules from Cryogenic Transmission Electron Microscope Images" In *Medical Imaging 2007: Physiology, Function, and Structure from Medical Images*, edited by Armando Manduca, Xiaoping P. Hu, Proc. of SPIE Vol. 6511. doi: 10.1117/12.710144
- Price, Catalina; Zhang, Jing; Wray, Keeley; Carroll, Mike; Panse, Anita; Peckham, Joan; Martin, Lenore M. (2008) "Universal Research Interchange (URI) Format: PDB to XML Format Converter" submitted to *Biopolymers: Peptide Science* 25 pages.
- Bradley T. Messmer, Lenore M. Martin, and David S. Thaler. (2008) "Monoclonal antibodies McPC603 and TEPC15 select different, non-crossreactive peptide analogs of phosphocholine from phage display libraries." Submitted to *Biopolymers*
- Lenore M. Martin, Sitaram Bhavaraju, and Bi-Huang Hu. (2008) "Oxazole, Thiazole, and Oxazoly-Thiazole Building Blocks Prepared from Amino Acids." Submitted to *Tetrahedron*

Books

- Martin, Lenore M.; Scovell, William (2003) *Student Study Guide & Problems Book for Campbell & Farrell's Biochemistry 4th ed.* Brooks/Cole, Thomson Learning Inc. 454 pages. ISBN 0-03-034917-6.
- Hu, Bi-Huang; Martin Lenore M. (1999) "Capillary Electrophoresis of Peptides and Proteins" Chap. 4 in *High-Resolution Chromatography: A Practical Approach*, Millner, P.A. Ed. IRL/Oxford Univ. Press; pp. 77-116. ISBN 0-19-963648-6.

Technical Reports

- URI-TR04-300 Martin, L.M.; Herve, J.Y.; Clayton, K.; Yun, H.; Lee, L.T. (2004) "URI Molecular Visualizer"
- URI-TR04-298 Peckham, J.; Martin, L.M. (2004) "Universal Research Interchange (URI) Format"

Patents

- Martin, Lenore M. and Hu, Bi-Huang (2000) "Oxazole and Thiazole Combinatorial Libraries"
US 09/936,972 Canada 2368026 EU 00919521.5 Japan 2000-606585

Manuscript and Book Review

- Regular Reviewer for *Journal of Antimicrobial Chemotherapy* 2002-present
- Regular Reviewer for *Bioorganic and Medicinal Chemistry Letters* 2006-present
- Ad-Hoc reviewer for *the Journal of Organic Chemistry*
- Ad-Hoc reviewer for *Tetrahedron Letters* and *Tetrahedron*
- Ad-Hoc reviewer and Book Reviewer for *the Journal of Medicinal Chemistry*
- Ad-Hoc reviewer for *Biopolymers*
- Ad-Hoc reviewer for *Letters in Peptide Science (LIPS)*
- Book reviews for *Drug Development and Industrial Pharmacy*

C. Research Support

Pending Research Support

(Martin, Collier, and Hervé) 4/1/2009-3/31/2014

PHS-NHLBI SF424 (R&R) PAR 07-344, Innovations in Biomedical Computer Science and Technology
"Bioinformatics Computer Vision Tools for Human Serum Lipoprotein Cryo-TEM Image Analysis"
Study how the human immune system detects subtle differences in the 3-D shapes of LDL particles in human serum, and gives distinct response profiles that may increase our understanding of pathways that lead to atherosclerosis when LDL concentrations are elevated. Develop a computer imaging algorithm that will be able to reliably determine the key 3-D shape parameters of LDL particles from only a few cryo-transmission electron microscopy (cryo-TEM) digital images without requiring hundreds of images as is required by the current single-particle reconstruction techniques.

Role: Lead PI

(DeGroot, Moise, Mather, Peckham, Paquette, Moss, Kornfeld, Gregory, Opal, & Martin) LOI 6/18/2008
PHS-NIAID U19 RFA-AI-03-015

Cooperative Centers for Translational Research on Human Immunology & Biodefense- **"Accelerating the Development of Immunome-Derived Vaccines for Emerging and Reemerging Infectious Disease"**
The immediate impact of the Martin Lab project will be to identify and test new families of antimicrobial peptides that can be further developed and used to combat the acute public health problem of rapidly emerging antibiotic resistance, in human pathogens. The long-term goal of our research is to develop an interdisciplinary collaboration with other researchers in the co-op that will generate a sustainable cross-pollination of ideas on how to advance the clinical management of rapidly-emerging drug-resistant human pathogens.

Role: PI of sub RO1 on Innate Immunity

(Greenfield and Martin) 7/1/2008-6/30/2013

NSF 07-603 Cyber-enabled Discovery and Innovation

"CDI-Type I: Modeling Side-Chain Packing in Midsize Proteins"

Natural proteins possess well-defined global free energy minima and hierarchical secondary and tertiary structure, all of which originate from the primary sequence of amino acids. One project objective is to understand how changes in side chain packing influence descriptors of the chain and/or chain fragments, both for a free chain and for a chain that can be perturbed by its environment, such as a membrane. Novel computational thinking will include devising appropriate coarse-grained and geometric measures for quantifying the shape, conformation, and polarity distribution of a chain and its side groups.

Role: Co-PI

Completed Research Support

(Cromarty, Martin, and Kass-Simon) 6/1/03-5/30/06

NSF

"Perception of the Molting Hormone, Twenty-hydroxy Ecdysone, and its Role in Orchestrating Aggressive Interactions in the American Lobster"

Use Western blots and capillary electrophoresis (CE) to identify and quantify the nuclear and membrane-bound 20-hydroxyecdysone receptor proteins and monitor their expression patterns in drosophila egg tissues, lobster eyestalks, and sensillae, so that protein expression can be correlated with behavioral criteria.

Role: Co-PI

(Gómez-Chiarri and Martin)

10/00-10/03

USDA-NRICGP Seed grant

"Feed-based Delivery of Recombinant Antimicrobial Peptides for Shellfish Aquaculture"

Design and chemical synthesis of antimicrobial peptides (AMPs) using solid-phase methods for prevention of vibriosis in aquaculture. Amphipathic alpha-helical peptide analogs of pleurocidin are being screened for selective activity against *Vibrio anguillarum*. Once AMPs with the desired characteristics are identified, active sequences will be expressed *in vivo*, cloned into oyster food stocks (algae) using genetic engineering.

Role: Co-PI

(Eckler and Martin)

2003

URI Provost's Office Undergraduate Research Proposal Award Winner

"Pleurocidin, A New Alternative to Antibiotics"

Antimicrobial peptides are promising substitutes for antibiotics. These peptides are found in skin cells and mucosal tissues in many vertebrates and invertebrates. The antimicrobial peptides act as natural host defense mechanisms against pathogenic bacteria, fungi, and viruses. One key advantage to using peptides composed of amino acids to develop new antibiotics is that the sequences can be easily modified whereas existing antibiotics are difficult to synthesize.

Role: Faculty Advisor

(Martin)

2000-2001

State of Rhode Island Forensic Partnership

"Capillary Electrophoresis for the Analysis of Nucleotides and DNA"

Investigation to determine the maximum sensitivity that can be obtained for analysis of DNA in blood samples using CE. Detailed method development, focusing on sample clean-up, buffers, and detector evaluation.

Role: PI

(Scapattici, Martin, and Kass-Simon)

5/00-5/02

Sigma Xi Grant-in-Aid and URI Foundation Competitive Grant

"The Biochemical Basis for Satiety in Hydra, a Fresh-Water Relative of Jelly-fish and Corals"

The project involves homogenizing Hydra prey and purifying an active component using bioassay-directed gel filtration, ion exchange, and HPLC. The purified substance was analyzed using mass spectrometry.

Role: Faculty Co-Sponsor

(Pray, Martin, and Hufnagel)

1998

URI Provost's Office Undergraduate Research Proposal Award Winner

"Isolation and Characterization of 'Fac', a Mating Factor from *Tetrahymena*"

Role: Faculty Sponsor

(Gandel et al.)

1998

NSF Meritorious Application Providers

"Computational and Network Intensive Applications in Biomedical and Molecular Research"

Role: Section 2.11, written by Martin and Nelson. (There were 13 sections, funding for infrastructure).

(Martin)

1995-1998

URI Foundation Competitive Grant

"A Gel Electrophoresis Laboratory for Pharmaceutical Analysis"

Role: PI

(Martin)

1996-1997

URI Foundation Proposal Development Award

"Synthesis and Evaluation of Model Membrane Proteins Based on Apolipoproteins"

Role: PI

(Martin)

1995-1996

URI Foundation Proposal Development Award

"Design, Synthesis and Screening of Combinatorial Libraries of DNA-Binding Antitumor Antibiotics"

Role: PI